284
A role for Notch signaling in the interpretation of cell fates in a morphogen gradient

Leonard Dobens, Benjamin Levine
Univ. Missouri-Kansas City, Kansas City, MO, USA

The morphogen concept has provided an enduring framework for understanding how tissue patterning occurs. According to current models, a morphogen molecule produced from a localized source of cells spreads to nearby cells to establish a signal gradient to direct alternative cell fates in a concentration-dependent manner. Experimental evidence from vertebrates and invertebrates indicates that TGFb family members act as morphogens and we use the *Drosophila* follicle cell (FC) epithelium as a model to understand how the TGFβ-related morphogen Dpp reproducibly generates discrete changes in gene expression and cell fate. During late oogenesis, DPP establishes (1) the anterior centripetal FC that produce the operculum and (2) the posterior columnar FC that produce the main body eggshell structure. In the centripetal FC, expression of the *bunched* (*bun*) gene is repressed at high levels of the DPP gradient. *bun*, which encodes a homolog of the mouse TSC-22/GILZ transcription factor, antagonizes Notch signaling to restrict centripetal FC fates. We present evidence that Notch directs expression of the C/EBP homolog *slow border cells* (*slbo*) in an initially broad group of cells at stage 9 and cross-repressive interactions among *slbo, bun* and Notch result in a sharp *slbo/bun* expression boundary by stage 10. The precise position of this boundary is sensitive to both Dpp and Notch levels and correlates with the position of the collar structure of the operculum. We propose that in FC exposed to a Dpp morphogen gradient, high and low levels of *slbo* and *bun*, respectively, are established by feedback to Notch signaling to direct threshold cell fates.

doi:10.1016/j.ydbio.2006.04.312

285
Tissue-specific regulation of DPP/BMP4 activity and signaling range by differential cleavage of the precursor in *Drosophila*

Shailaja Sopory, Marcel Wehrli, Jan L. Christian
Oregon Health and Science Univ., Portland, OR, USA

BMP4 is sequentially cleaved at an optimal furin motif (S1), followed by cleavage at a minimal furin motif (S2) within the prodomain, which regulates the activity and signaling range of BMP4 ligand. We have utilized *Drosophila* as a model system to determine whether differential cleavages within the prodomain of BMP4/DPP provide an evolutionarily conserved mechanism for regulating their activity in a tissue-specific fashion and also to visualize and compare the morphogen gradient formed by wild-type and an S2 cleavage mutant form of DPP. DPP is known to form long- and short-range signaling gradients in the wing disc and midgut, respectively, thus providing a good system to test the question of tissue-specific cleavage. We made tagged versions of DPP, characterized them in vitro, generated transgenic flies harboring the wild-type (UAS HA DPP-Myc^{WT}) and S2 cleavage mutant (UAS HA DPP-Myc^{S2KK}) form of DPP, and looked at the ability of DPP-Myc cleaved from wild-type and mutant precursor to rescue wing formation in flies with the genotype $dpp^{d8}/dppd^{10}$. Adults of this genotype have smaller eyes and reduced or absent wings. Expression was driven by GAL4 under control of a *dpp* promoter. DPP-Myc cleaved from wild-type precursor rescued the reduced wing and eye phenotype, whereas that cleaved from the proHA DPP-Myc^{S2KK} precursor rescued the small eye phenotype but not wing formation. These observations support our hypothesis that cleavage at both sites is required to generate a long-range signaling gradient in the wing disc and further suggest that DPP signaling in the eye requires cleavage at only the S1 site.

doi:10.1016/j.ydbio.2006.04.313

286
Delayed effect of a maternal BMP on establishment of the embryonic dorsal–ventral axis

Helena M. Araujo [1], Katia Carneiro [1], Marcio R. Fontenele [1], Erika M. Negreiros [1], Ethan Bier [2]
[1] *Federal University of Rio de Janeiro, Rio de Janeiro, Brazil*
[2] *University of California at San Diego, La Jolla, CA, USA*

The dorsal–ventral (DV) axis of the *Drosophila* embryo depends on establishment of a gradient of the NFκB/c-rel-related transcription factor dorsal. Two pathways have been proposed to modulate formation of this gradient, based on controlling the levels of the IκB homologue cactus. Signaling through the Toll receptor induces degradation of cactus, releasing dorsal for nuclear translocation. Signaling through maternal BMP increases the amount of cactus, retaining dorsal in the cytoplasm. The BMP4 homologue *decapentaplegic* (*dpp*) and the BMP antagonist encoded by *short gastrulation* (*sog*) are expressed by follicle cells during mid-oogenesis, performing roles in DV patterning of both eggshell and embryo. Here we explore the mechanism that enables these proteins to perform this dual role. We provide evidence that maternal Sog and Dpp proteins are secreted into the perivitelline space where they remain until early embryogenesis to modulate cactus degradation. We find that metalloproteases encoded by *tolloid* (*tld*) and *tolkin* (*tok*), which cleave Sog, are expressed by follicle cells and are required to generate DV asymmetry in the Dpp signal. Examination of embryonic DV expression territories suggests that maternal Dpp modifies the slope of the dorsal gradient. Since expression of *tld* and *tok* is ventrally restricted by the TGF-α ligand encoded by *gurken*, we propose that the EGFR receptor pathway assures correct establishment of embryonic DV territories by spatially regulating both Toll and BMP signaling.

doi:10.1016/j.ydbio.2006.04.314